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(54) Title: COMPOSITION AND METHOD OF PREPARING MICROPARTICLES OF WATER–INSOLUBLE SUBSTANCES

(57) Abstract

Compositions and procedures that yield sub–micron and micron–size stable particles of water–insoluble or poorly soluble drugs or other industrially useful insoluble compounds are prepared using combinations of natural or synthetic phospholipids, a charged surface modifier such as a highly purified charged phospholipid and a block copolymer coated or adhered onto the surfaces of the water insoluble–compound particles. The combination of charged surface modifier and block copolymer allows the formation and stabilization of the sub–micron and micron size compound particles – stabilized by the charged phospholipid to provide electrostatic stabilization and the block copolymer to provide steric stabilization – and therefore prevents these particles from particle growth, aggregation or flocculation.

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COMPOSITION AND METHOD OF PREPARING MICROPARTICLES OF WATER-INSOLUBLE SUBSTANCES

This application claims benefit of provisional application Serial No. 60/079,809 filed March 30, 1998, the disclosure of which is hereby incorporated by reference.

This invention relates to compositions and procedures that yield sub-micron and micron-size stable particles of water-insoluble or poorly soluble drugs or other industrially useful insoluble compounds. The compositions of this invention include combinations of natural or synthetic phospholipids, a charged surface modifier such as a highly purified charged phospholipid and a block copolymer coated or adhered onto the surfaces of the water insoluble-compound particles. The combination of charged surface modifier and block copolymer allows the formation and stabilization of the sub-micron and micron size compound particles--stabilized by the charged phospholipid surface modifiers to provide electrostatic stabilization and the block copolymer to provide steric stabilization--and therefore prevent these particles from particle growth, aggregation or flocculation.

BACKGROUND OF THE INVENTION

There is a critical need in the pharmaceutical and other biological based industries to formulate water-insoluble or poorly soluble substances into formulations for oral, injectable, inhalation and ophthalmic routes of delivery. Water insoluble compounds are those having poor solubility in water, that is < 5 mg/ml at physiological pH (6.5-7.4). Preferably their water solubility is < 1 mg/ml, more preferably < 0.1 mg/ml. It is desirable that the drug is stable in water as a dispersion; otherwise a lyophilized or spray-dried solid form may be desirable.

As used herein, "micro" refers to a particle having diameter of from nanometers to micrometers. Microparticles, as used herein, refer to solid particles of irregular, non-spherical or spherical shapes. Formulations containing these microparticles provide some specific advantages over the unformulated non-micronized drug particles, which include improved oral bioavailability of drugs that are poorly absorbed from GI tract, development of injectable formulations that are currently available only in oral dosage form, less toxic injectable formulations that are currently prepared with organic solvents, sustained release of intramuscular injectable drugs that are currently administered through daily injection or

constant infusion, and preparation of inhaled, ophthalmic formulation of drugs that otherwise could not be formulated for nasal or ocular use.

Current technology for delivering insoluble drugs as described in US Patents 5,091,188; 5,091,187 and 4,725,442 focuses on (a) either coating small drug particles with natural or synthetic phospholipids or (b) dissolving the drug in a suitable lipophilic carrier and forming an emulsion stabilized with natural or semisynthetic phospholipids. One of the disadvantages of these formulations is that certain drug particles in suspension tend to grow over time because of the dissolution and reprecipitation phenomenon known as the "Oswald ripening" or particle growth, as the solvent becomes saturated with solute, the larger particles grow and become even larger, Luckham, Pestic. Sci., (1999) **25**, 25-34.

Another approach, as described in a series of patents uses a cloud point modifier(s). In U.S. 5,298,262; 5,326,552; 5,336,507; 5,304,564 and 5,470,583 a poorly soluble drug or diagnostic agent has adsorbed on its surface both a cloud-point modifier and a non-crosslinked nonionic surfactant. The role of the cloud point modifier is to increase the cloud point of the surfactant such that the resulting nanoparticles are resistant to particle size growth upon heat sterilization at 121°C.

DESCRIPTION OF THE INVENTION

The present invention focuses on preparing submicron to micron size particles using a combination of electrostatic and steric stabilization using at least one charged surface modifier and at least one block copolymer, with particles coated with a natural phospholipid. In this manner the growth of particle size, and hence storage stability, is controlled by adding a combination of electrostatic and steric stabilizing materials.

The use of this particular combination of electrostatic and steric stabilizers in addition to a natural phospholipid is characterized by its ability to result in volume weighted mean particle size values that are smaller than what can be achieved using phospholipid alone without the use of a surfactant with the same energy input, and provide compositions resistant to particle size growth on storage. In order to achieve the advantages of the present invention it is necessary that the natural phospholipid and stabilizers all be present at the time of particle size reduction or precipitation.

Another aspect of the present invention includes free-flowing powders of poorly soluble or insoluble drug substances such as cyclosporin as well as solid dosage forms of these powders, for instance in the form of compressed tablets and the like. Surprisingly we have found that microparticle formulations exhibit enhanced stability as illustrated in the data that follows.

Although we do not wish to be bound by any particular theory, it appears that these surface modifiers generally, that is phospholipids and one or more surfactants, adsorb to the surfaces of drug particles, and (a) convert lipophilic to hydrophilic surfaces with increased steric hindrance/stability, and (b) possibly modify zeta potential of surfaces with more charge repulsion stabilization. The concentrations of surface modifiers used in the process described here are normally above their critical micelle concentrations (CMC) and hence facilitate the formation of sub-micron to micron particles by stabilizing the small particles as they are formed to prevent reaggregation.

Phospholipid and surface modifier(s) are adsorbed onto the surfaces of drug particles in sufficient quantity to retard drug particle growth, reduce drug average particle size from 5 to 100 μ to sub-micron and micron size particles by one or combination of methods known in the art, such as sonication, homogenization, milling, microfluidization, precipitation or recrystallization or precipitation from supercritical fluid, and maintain sub-micron and micron size particles on subsequent storage as suspension or solid dosage form.

The formulations prepared by this invention may be dried, e.g., by lyophilization, fluid or spray drying, into powders, which can be resuspended or filled into capsules or converted into granules or tablets with the addition of binders and other excipients known in the art of tablet making.

By industrially useful insoluble or poorly soluble compounds we include biologically useful compounds, imaging agents, pharmaceutically useful compounds and in particular drugs for human and veterinary medicine. Water insoluble compounds are those having a poor solubility in water, that is less than 5 mg/ml at a near neutral pH of 5 to 8, although the water solubility may be less than 1 mg/ml and even less than 0.1 mg/ml.

Examples of some preferred water-insoluble drugs include immunosuppressive agents such as cyclosporins including cyclosporine (cyclosporin A), immunoactive agents, antiviral

and antifungal agents, antineoplastic agents, analgesic and anti-inflammatory agents, antibiotics, anti-epileptics, anesthetics, hypnotics, sedatives, antipsychotic agents, neuroleptic agents, antidepressants, anxiolytics, anticonvulsant agents, antagonists, neuron blocking agents, anticholinergic and cholinomimetic agents, antimuscarinic and muscarinic agents, antiadrenergic and antiarrhythmics, antihypertensive agents, hormones, and nutrients. A detailed description of these and other suitable drugs may be found in *Remington's Pharmaceutical Sciences*, 18th edition, 1990, Mack Publishing Co. Philadelphia, PA.

The phospholipid may be any naturally occurring phospholipid or mixtures of phospholipids, sometimes referred to herein as "commercial" phospholipids, such as egg or soybean phospholipid or a combination thereof. The phospholipid may be salted or desalted, hydrogenated or partially hydrogenated or natural semisynthetic or synthetic. Examples of commercially available phospholipids include but are not limited to egg phospholipids P123 (Pfanstiehl), Lipoid E80 (Lipoid); and hydrogenated soy phospholipids Phospholipon 90H and 100H (Natterman) and 99% pure egg and soy phosphatidyl choline (Avanti Polar Lipids). The amount of phospholipid present in the composition ranges from 0.01% to 50%, preferably from 0.05% to 20%.

Block copolymers used in the invention display a brush-like interfacial conformation and possible steric stabilization to the particles. Suitable block copolymers include polaxomers, such as Pluronic™ F68, F108 and F127, which are block copolymers of ethylene oxide and propylene oxide available from BASF; and poloxamines, such as Tetronic™ 908 (T908, T707, T909, T1107 and T1307), which are tetrafunctional block copolymers derived from sequential addition of ethylene oxide and propylene oxide to ethylene-diamine available from BASF. In a preferred aspect of the invention, when free-flowing formulations are desired, the block copolymer will itself be a powder. The amount of block copolymer is between 0.01% and 20%, preferably from 0.1% to 10%.

The charged surface modifier(s) used in the present invention are highly purified phospholipids either isolated from natural products or prepared synthetically. For example, commercially available phosphatidylcholine contains a small percentage of charged phosphatides such as phosphatidyl glycerol, phosphatidyl inosite, phosphatidyl serine and phosphatidic acid and its salts. Other charged phospholipids include palmitoyl-oleyl-

phosphatidyl-glycerol (POPG) and dimiristoyl phosphatidylglycerol sodium salt (DMPG). Combinations of charged phospholipids may be used. These materials are present in relatively small amounts and serve to allow smaller particle formation and inhibit aggregation. The amount of charged phospholipids ranges from 0.01% to 5.0% and preferably from 0.05% to 1.0%.

It is thought that some of the functions of the combination of surface modifiers as it relates to this invention are (a) suppressing the process of Oswald Ripening and therefore maintaining the particle size, (b) increasing the storage stability, minimizing agglomeration and sedimentation, and decreasing the particle growth during lyophilization and reconstitution; (c) adhering or coating firmly onto the surfaces of water-insoluble drug particles and therefore modifying the interfaces between the particles and the liquid in the resulting formulations; (d) increasing the interface compatibility between water-insoluble drug particles and the liquid; and (e) possibly orienting preferentially themselves with the hydrophilic portion sticking into the aqueous solution and the lipophilic portion strongly adsorbed at the water-insoluble drug particle surfaces; and (f) preventing aggregation of the small particles back to larger particles as they are being formed using size reducing equipment or precipitation.

Considerable variations as to the identities and types of charged surface modifier and especially the block copolymer should be expected depending upon the drug or active agent selected as the surface properties of these small particles are different. The most advantageous agents for the insoluble drug will be apparent following empirical tests to identify the system/combination resulting in the requisite particle size and particle size stability on storage over time.

Various procedures can be used to produce these stable sub-micron and micron size particles including mixing the insoluble substance with charged surface modifier and block copolymer followed by sonication, milling, homogenization, microfluidization; or precipitating from a solution of the substance using antisolvent and solvent precipitation in the presence of the phospholipid and surfactant(s). Mannitol and other disaccharides and other agents may be added to adjust the final formulation to isotonicity as well as acting as a stabilizing aid during drying.

Unless otherwise specified, all parts and percentages reported herein are weight per unit volume (w/v), in which the volume in the denominator represents the total volume of the system. Diameters of dimensions are given in millimeters (mm = 10^{-3} meters), micrometers (μm = 10^{-6} meters), nanometers (nm = 10^{-9} meters) or Angstrom units (= 0.1 nm). Volumes are given in liters (L), milliliters (mL = 10^{-3} L) and microliters (μL = 10^{-6}L). Dilutions are by volume. All temperatures are reported in degrees Celsius. The compositions of the invention can comprise, consist essentially of or consist of the materials set forth and the process or method can comprise, consist essentially of or consist of the steps set forth with such materials.

The invention is further explained with reference to the following preferred embodiments. The following general procedure was used for the examples; exceptions are noted.

Preparation of premix. Commercial phospholipid, mannitol, charged surface modifier and block copolymer were first mixed with water using a hand mixer. The drug was added afterwards to the mixture, and mixed for 10 min-30 min at room temperature. In the case of cyclosporine, the pH was adjusted to 7.5-8.0 using 1N NaOH, and the premix was cooled to 12°C using an ice bath. The batch size for cyclosporin was 200 g, for ursodiol 50 g and for fenofibrate 200 g.

Processing conditions. The premix was processed at a constant temperature and pressure by using high-pressure instrumentation that subjects the formulation to shear, cavitation, impact, and attrition, that is in either a microfluidizer or a homogenizer.

Formulation type	Processing Machine	Total Passes at Operating Pressure	Average Pressure (kPsi)	Average Temper-ature (C)
Cyclosporine	Avestin C-50 homogenizer	200	18	10
Ursodiol	Avestin C-5 homogenizer	100	18	13
Fenofibrate	Microfluidizer M110H	50	18	5

A "pass" is defined as one cycle of the formulation through the different elements of the processing machine. The "pass" or cycle for each machine is as follows:

Avestin C-50 and C-5: Formulation is placed in inlet reservoir then passes to the homogenization valve, next a heat exchanger then back to the inlet reservoir. It is the homogenization valve that subjects the formulation to the forces of shear, cavitation, impact and attrition.

M110H: The formulation is first put through 20 passes of the bypass loop, defined as follows: inlet reservoir to auxiliary processing module to heat exchanger then back to inlet reservoir. The resulting formulation is then put through the interaction chamber loop, defined as follows: inlet reservoir to auxiliary processing module to interaction chamber to heat exchanger then back to inlet reservoir. it is in the interaction chamber where the formulation is subject to the forces of shear, cavitation, impact and attrition. Following processing, each formulation was collected and placed in vials for stability testing. "MP" indicates microparticles falling within the range of 0.05 to 10 microns.

The five different types of stability tests are described as follows:

Stability Test	Description
4C	Sample stored at 4°C (temperature controlled)
25C	Sample stored at 25°C (temperature controlled, 60% relative humidity)
25C(2)	Sample stored at ambient room temperature -cyclosporine only
40C	Sample stored at 40°C (temperature controlled)
Shaking	Sample laid down on its side on a shaking table at ambient room temperature. The shaking speed was at 100 rpm-110 rpm.
Thermal Cycling	One cycle defined as follows: sample stored at 4°C for 2-4 days, then at 40°C for 2-4 days.

EXAMPLE 1

Effect of steric and charged surface modifiers on particle size reduction. These experiments show that in the presence of phospholipid a combination effect of steric and charged stabilizers gives a smaller terminal particle size, than by using either alone. In all cases, the total weight percent of surface modifiers (commercial phospholipid, block copolymer, charged surface modifier) is kept constant.

Table 1.1 – MP Cyclosporine data (5% w/w micronized cyclosporine, 5.5% Mannitol)
200 g batches, processed on homogenizer Avestin C-50

Sample	w/w% Lipoid E80	w/w% DMPG	w/w% Pluronic F68	# passes	Particle size (microns)
1	10	0	0	209	2.62
2	9.5	0.5	0	217	1.20
3	9.0	0	1.0	177	1.77
4	8.7	0.45	0.95	210	1.08

Table 1.2 – MP Ursodiol data (10% w/w Ursodiol, 5.5% Mannitol)
50g batches, processed on homogenizer Avestin C-5

Sample	w/w% Lipoid E80	w/w% DPPE	w/w% Tetronic 908	# passes	Particle size (microns)
1	2.4	0	0	127	1.36
2	1.6	0	0.8	107	1.15
3	2.0	0.4	0	106	1.34
4	1.41	0.28	0.72	102	1.06
5	0	0.4	2.0	104	1.37

Table 1.3 – MP Fenofibrate data (10% w/w Fenofibrate, 5.5% Mannitol)
200g batches, processed on Microfluidizer M110H

Sample	w/w% Lipoid E80	w/w% DMPG	w/w% Poloxamer 407	# passes	Particle size (microns)
1	4.0	0	0	70	0.95
2	3.0	0	1.0	70	0.86
3	3.6	0.4	0	70	0.85
4	2.77	0.31	0.92	70	0.82

The data for cyclosporine, ursodiol and fenofibrate show the particle size reduction is maximal in phospholipid coated microparticles in the presence of charged surface modifier and a block copolymer.

EXAMPLE 2

Effect of the presence of steric and charged stabilizers on the rate of particle size reduction. As the formulation passes through the homogenizer, the average diameter of the formulated particles reduces in magnitude. An empirical relation has been found that relates the average diameter to the pass number:

$$\text{Average diameter} = K/(\text{pass number})^{a}$$

The above equation can also be used to determine how many passes it takes for the average diameter to reduce to 1 micron: # of passes to reach 1 micron = $(K)^{1/a}$. These data demonstrate that steric and charged stabilizers improve the rate of particle formation.

Table 2.1 – MP Fenofibrate data (10% w/w Fenofibrate, 5.5% Mannitol)
Rate of particle size reduction - 200g batches on the Microfluidizer M110H

Sample	w/w% Lipoid E80	w/w% DMPG	w/w% Poloxamer 407	Calculated # passes for 1 micron*
1	4.0	0	0	44
2	3.0	0	1.0	33
3	3.6	0.4	0	37
4	2.77	0.31	0.92	27

*For Fenofibrate, the total pass number is the calculated value plus 20 passes of the formulation using the bypass loop.

Table 2.2 – MP Ursodiol data (10% w/w Ursodiol, 5.5% Mannitol)
Rate of particle size reduction - 50g batches on the Avestin C5 homogenizer

Sample	w/w% Lipoid E80	w/w% DPPE	w/w% Tetronic 908	Calculated # passes for 1 micron
1	2.4	0	0	305
2	1.6	0	0.8	158
3	2.0	0.4	0	261
4	1.41	0.28	0.72	134
5	0	0.4	2.0	230

The data for ursodiol and fenofibrate show the rate of particle size reduction is maximal in the production of phospholipid coated microparticles in the presence of charged surface modifier and a block copolymer.

EXAMPLE 3

Effect of steric and charged surface modifiers on particle stability. These data demonstrate the combination of charged phospholipid and block copolymer provide stability against Ostwald ripening and aggregation of the particles in the formulations.

Cyclosporine

Table 3.1 – MP Cyclosporine data (5% w/w micronized cyclosporine, 5.5% Mannitol)
Particle size at room temperature

Sample	w/w% Lipoid E80	w/w% DMPG	w/w% Pluronic F68	Initial size (microns)	Final size (microns)	Days
1	10	0	0	2.62	8.07	66
2	9.5	0.5	0	1.20	1.64	61
3	9.0	0	1.0	1.77	6.74	53
4	8.7	0.45	0.95	1.08	1.24	51

The combination of effect of steric and electrostatic stabilizers provides best stability and prevents or minimizes particle growth due to both Ostwald ripening and particle aggregation.

Ursodiol

Table 3.2 – MP Ursodiol data (10% w/w Ursodiol, 5.5% Mannitol)
Particle size at 4 C

Sample	w/w% Lipoid E80	w/w% DPPE	w/w% Tetronic 908	Initial size (microns)	Final size (microns)	Days
1	2.4	0	0	1.36	1.52	30
2	1.6	0	0.8	1.15	1.20	29
3	2.0	0.4	0	1.34	1.33	27
4	1.41	0.28	0.72	1.06	1.13	26
5	0	0.4	2.0	1.37	1.34	13

Table 3.3 – MP Ursodiol data (10% w/w Ursodiol, 5.5% Mannitol)
Particle size at room temperature

Sample	w/w% Lipoid E80	w/w% DPPE	w/w% Tetronic 908	Initial size (microns)	Final size (microns)	Days
1	2.4	0	0	1.36	1.51	30
2	1.6	0	0.8	1.15	1.19	29
3	2.0	0.4	0	1.34	1.55	29
4	1.41	0.28	0.72	1.06	1.13	26
5	0	0.4	2.0	1.37	1.44	24

Table 3.4 – MP Ursodiol data (10% w/w Ursodiol, 5.5% Mannitol)
Particle size at 40C

Sample	w/w% Lipoid E80	w/w% DPPE	w/w% Tetronic 908	Initial size (microns)	Final size (microns)	Days
1	2.4	0	0	1.36	1.51	30
2	1.6	0	0.8	1.15	1.23	29
3	2.0	0.4	0	1.34	1.35	27
4	1.41	0.28	0.72	1.06	1.12	26
5	0	0.4	2.0	1.37	1.35	20

Table 3.5 – MP Ursodiol data (10% w/w Ursodiol, 5.5% Mannitol)
Shaking stability data at room temperature

Sample	w/w% Lipoid E80	w/w% DMPG	w/w% Tetronic 908	Initial size (microns)	Final size (microns)	Days
1	2.4	0	0	1.36	-	-
2	1.6	0	0.8	1.15	1.17	15
3	2.0	0.4	0	1.34	1.36	7
4	1.41	0.28	0.72	1.06	1.09	7
5	0	0.4	2.0	1.37	1.37	7

Table 3.6 – MP Ursodiol data (10% w/w Ursodiol, 5.5% Mannitol)
Thermal cycling stability data (after 3 cycles)

Sample	w/w% Lipoid E80	w/w% DMPG	w/w% Tetronic 908	Initial size (microns)	Final size (microns)
1	2.4	0	0	1.36	-
2	1.6	0	0.8	1.15	1.21
3	2.0	0.4	0	1.34	1.36
4	1.41	0.28	0.72	1.06	1.13

The sample (example 4) prepared with a combination of electrostatic and steric surface modifiers showed good stability under all conditions.

Fenofibrate

Table 3.7– MP Fenofibrate data (10% w/w Fenofibrate, 5.5% Mannitol)
Particle size at 4C

Sample	w/w% Lipoid E80	w/w% DMPG	w/w% Poloxamer 407	Initial size (microns)	Final size (microns)	Days
1	4.0	0	0	0.95	3.59	30
2	3.0	0	1.0	0.86	1.10	33
3	3.6	0.4	0	0.85	2.91	33
4	2.77	0.31	0.92	0.82	1.17	32

Table 3.8– MP Fenofibrate data (10% w/w Fenofibrate, 5.5% Mannitol)
Particle size at 25C

Sample	w/w% Lipoid E80	w/w% DMPG	w/w% Poloxamer 407	Initial size (microns)	Final size (microns)	Days
1	4.0	0	0	0.95	6.47	30
2	3.0	0	1.0	0.86	1.32	29
3	3.6	0.4	0	0.85	8.10	29
4	2.77	0.31	0.92	0.82	1.39	28

Table 3.9 – MP Fenofibrate data (10% w/w Fenofibrate, 5.5% Mannitol)
Shaking stability data

Sample	w/w% Lipoid E80	w/w% DMPG	w/w% Poloxamer 407	Initial size (microns)	Final size (microns)	Days
1	4.0	0	0	0.95	3.53	8
2	3.0	0	1.0	0.86	1.27	7
3	3.6	0.4	0	0.85	2.86	7
4	2.77	0.31	0.92	0.82	1.32	7

Table 3.10 – MP Fenofibrate data (10% w/w Fenofibrate, 5.5% Mannitol)
Thermal cycling stability data

Sample	w/w% Lipoid E80	w/w% DMPG	w/w% Poloxamer 407	Initial size (microns)	Final size (microns)	# cycles
1	4.0	0	0	0.95	3.59	3
2	3.0	0	1.0	0.86	2.26	3
3	3.6	0.4	0	0.85	8.61	3
4	2.77	0.31	0.92	0.82	2.54	3

The sample (example 4) prepared with a combination of charged and steric surface modifiers showed good stability under all conditions.

The presence of a charged and steric surface modifiers during the formation of micron to sub-micron sized phospholipid coated microparticles provides for the high rate of production of minimally sized particles. Also, the combination of effect of steric and electrostatic stabilizers provides best stability and prevents or minimizes particle growth due to both Ostwald ripening and particle aggregation. Further, charged surface modifiers appear possibly to contribute mostly to particle size reduction whereas steric modifiers contribute mostly to stability.

The above data demonstrate the presence of a charged and steric surface modifiers during the formation of micron to sub-micron sized phospholipid coated microparticles provides for a high rate of production of minimally sized particles.

The following materials were used in the above examples:

TABLE 1 – Surface Modifiers

Full Name	**Abbreviation**	**Class of surface modifier**	**Type of stabilization**
Lipoid E-80	LipE80	Phospholipid	
1,2-Dimyristoyl-sn-Glycero-3-[Phospho-rac-(1-glycerol)](Sodium Salt)	DMPG	Charged	Electrostatic
1,2-Dipalmitoyl-sn-Glycero-3-Phosphoethanolamine	DPPE	Charged	Electrostatic
Pluronic F127 (also known as Poloxamer 407)	PF127	Block Copolymer	Steric
Tetronic 908	T908	Block Copolymer	Steric
Pluronic F68 (also known as Poloxomer 188)	PF68	Block copolymer	Steric

TABLE 2 – List of Suppliers		
Name	**Supplier**	**Location**
Cyclosporine	North China Pharmaceutical Company	China
Ursodiol	Tokyo Tanabee	Tokyo, Japan
Fenofibrate	Laboratorio Chimico Internazionale s.p.a.	Milan, Italy
Lipoid E-80	Lipoid GMBH	Ludwigshafen, Germany
DMPG, DPPE	Avanti Polar Lipids	Alabaster, Alabama, USA
Tetronic and Pluronic Block Polymers	BASF	Mount Olive, New Jersey, USA

WHAT IS CLAIMED IS:

1. A pharmaceutical composition comprising particles having diameters of about 0.05 to about 10 microns of a water-insoluble or poorly soluble drug having phospholipid coated surfaces of the drug particles stabilized with a charged surface modifier and a block copolymer wherein the charged surface modifier provides electrostatic stabilization and the block copolymer provides steric stabilization against Ostwald ripening and particle aggregation and provides for small particle formation

2. A pharmaceutical composition comprising particles having diameters of about 0.05 microns to about 10 microns of a water-insoluble or poorly soluble drug, the particles coated with a mixture of 0.01% to 50% wt naturally occurring phospholipids, 0.01 to 5.0% wt of a charged surface modifier and 0.01 to 20% wt of a block copolymer, wherein the charged surface modifier provides electrostatic stabilization and the block copolymer provides steric stabilization against Ostwald ripening and particle aggregation and provides for small particle formation.

3. The composition of claims 1 and 2 wherein the block copolymer is derived from ethylene oxide and propylene oxide, or a tetrafunctional block copolymer derived from sequential addition of ethylene oxide and propylene oxide to ethylene diamine or combinations thereof.

4. The composition of claim 1 or 2 wherein the phospholipid is a phospholipid of egg or plant origin or semisynthetic or synthetic in partly or fully hydrogenated form or in a desalted or salt form.

5. The composition of claim 2 wherein the naturally occurring phospholipid is present in an amount of 0.05% to 20% wt.

6. The composition of claim 1 or 2 wherein the charged surface modifier is selected from the charged phospholipids phosphatidylcholine, dimyristoyl phosphatidylglycerol sodium salt, phosphatidylethanolamine, phosphatidylserine, phosphatidic acid, or combinations thereof.

7. The composition of claim 6 wherein the charged surface modifier is present in an amount of 0.05% to 1.0% wt.

8. The composition of claim 3 wherein the block copolymer is present in an amount of 0.1% to 10%.

9. The pharmaceutical composition of claim 1 or 2 in sterile, injectable form for intravenous, intra-arterial, intra-muscular, intradermal, subcutaneous intra-articular, cerebrospinal, epidural, intracostal, intraperitoneal, intratumor, intrabladder, intra-lesion or subconjunctival administration.

10. A suspension, spray-dried powder, lyophilized powder granules or tablets of the composition of claim 1 or 2.

11. A hard or soft gel capsule formulation comprising the composition of claim 1 or 2.

12. A method of increasing the rate of microparticle formation of a pharmaceutical composition comprising particles of a water-insoluble or poorly soluble drug having phospholipid coated surfaces, said method comprising homogenizing or microfluidizing to produce the phospholipid coated drug particles in the presence of a charged surface modifier and a block copolymer, wherein the charged surface modifier provides electrostatic stabilization and the block copolymer provides steric stabilization against Ostwald ripening and particle aggregation resulting in particles having a diameter of about 0.05 to about 10.

13. A method of increasing the rate of particle formation of a pharmaceutical composition comprising particles of a water-insoluble or poorly soluble drug, comprising homogenizing or microfluidizing the particles in the presence of a mixture of 0.01% to 50% wt naturally occurring phospholipids, 0.01 to 5.0% wt of charged surface modifier and 0.01 to 20% wt of a steric stabilizing block copolymer, wherein the charged surface modifier provides electrostatic stabilization and the block copolymer provides steric stabilization against Ostwald ripening and particle aggregation resulting in particles having diameters of about 0.05 microns to about 10 microns.

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(81) Designated States: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

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18 November 1999 (18.11.99)

(54) Title: COMPOSITIONS CONTAINING MICROPARTICLES OF WATER-INSOLUBLE SUBSTANCES AND METHOD FOR THEIR PREPARATION

(57) **Abstract**

Compositions and procedures that yield sub–micron and micron–size stable particles of water–insoluble or poorly soluble drugs or other industrially useful insoluble compounds are prepared using combinations of natural or synthetic phospholipids, a charged surface modifier such as a highly purified charged phospholipid and a block copolymer coated or adhered onto the surfaces of the water insoluble–compound particles. The combination of charged surface modifier and block copolymer allows the formation and stabilization of the sub–micron and micron size compound particles – stabilized by the charged phospholipid to provide electrostatic stabilization and the block copolymer to provide steric stabilization – and therefore prevents these particles from particle growth, aggregation or flocculation.

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INTERNATIONAL SEARCH REPORT

International Application No
PCT/US 99/06746

A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 A61K9/51 A61K9/16 A61K38/13 A61K31/575 A61K31/215
A61K47/10 A61K47/24

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 6 A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 98 07414 A (RES TRIANGLE PHARMACEUTICALS L) 26 February 1998 (1998-02-26) abstract page 1, line 4 - line 14 page 2, line 26 - page 4, line 17 page 5, line 12 - page 6, line 14 page 7, line 4 - line 9 examples 1,2 claims 1-17 --- -/--	1-13

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

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"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
20 July 1999	29 September 1999

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Taylor, G.M.

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Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

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PCT/US 99/06746

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 97 14407 A (RES TRIANGLE PHARMACEUTICALS ;UNIV TEXAS (US); HENRIKSEN INGE B (U) 24 April 1997 (1997-04-24) abstract page 1, line 20 - page 2, line 2 page 3, line 10 - line 27 page 4, line 17 - page 6, line 18 page 10, line 3 - line 14 example 1 claims 1-17	1-13
X	GUZMAN M: "FORMATION AND CHARACTERIZATION OF CYCLOSPORINE-LOADED NANOPARTICLES" JOURNAL OF PHARMACEUTICAL SCIENCES, vol. 82, no. 5, 1 May 1993 (1993-05-01), pages 498-502, XP000368107 ISSN: 0022-3549 abstract	1-13
X	EP 0 499 299 A (STERLING WINTHROP INC) 19 August 1992 (1992-08-19) abstract page 3, line 19 - line 53 page 4, line 16 - line 47 claims 1-20	1-13
A	WO 96 24332 A (NANOSYSTEMS LLC) 15 August 1996 (1996-08-15) abstract page 1, line 5 - line 14 examples 1-4 claims 1-6	1-13
E	WO 99 29300 A (MISHRA AWADHESH K ;PARIKH INDU (CA); MOUSSA ISKANDAR (CA); RTP PHA) 17 June 1999 (1999-06-17) abstract page 5, line 19 - page 6, line 3 examples 1-14 claims 1-18	1-13

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INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 99/06746

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☒ Claims Nos.: 1,2,12,13
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
see FURTHER INFORMATION sheet PCT/ISA/210

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest.

☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.2

Claims Nos.: 1,2,12,13

Present claims 1, 2, 12 and 13 relate to a product/method defined by reference to a desirable characteristic or property, namely that

"the charged surface modifier provides electrostatic stabilisation and the block copolymer provides steric stabilisation against Ostwald ripening and particle aggregation and provides for small particle formation"

The claims cover all products/methods having this characteristic or property, whereas the application provides support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT for only a very limited number of such products/methods. In the present case, the claims so lack support, and the application so lacks disclosure, that a meaningful search over the whole of the claimed scope is impossible.

Independent of the above reasoning, the claims also lack clarity (Article 6 PCT). An attempt is made to define the product/method by reference to a result to be achieved. Again, this lack of clarity in the present case is such as to render a meaningful search over the whole of the claimed scope impossible. Consequently, the search has been carried out for those parts of the claims which appear to be clear, supported and disclosed, namely those parts relating to the products/methods, namely

the block co-polymers as defined in claim 3, and

the charged surface modifiers as defined in claim 6.

The applicant's attention is drawn to the fact that claims relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/US 99/06746

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